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8: Calderone R, Diamond R, Senet JM, Warmington J, Filler S, Edwards JE.	Related Articles Links
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☐ 10: Franklyn KM, Warmington JR, Ott AK, Ashman RB. An immunodominant antigen of Candida albicans shows homo enolase. Immunol Cell Biol. 1990 Jun;68 (Pt 3):173-8. PMID: 2228032 [PubMed - indexed for MEDLINE]	Related Articles, Links logy to the enzyme
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Feb 21, 1989

US-PAT-NO: 4806465

DOCUMENT-IDENTIFIER: US 4806465 A

TITLE: Cytoplasmic antigens of candida albicans and methods of using the same

DATE-ISSUED: February 21, 1989

INVENTOR - INFORMATION:

NAME

CITY

STATE ZIP CODE

COUNTRY

Buckley; Helen R.

Philadelphia

PA

Philadelphia PA

Largen; Michael T. Strockbine; Nancy A.

Bethesda

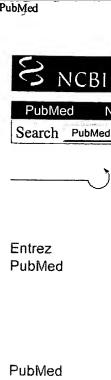
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CLAIMS:

What is claimed is:

- 1. A diagnostic method for disseminated or invasive candidiasis comprising:
- a. contacting blood serum with a composition containing a substantially bichemically pure preparation of a <u>cytoplasmic</u> antigen of C. albicans, said antigen having an apparent molecular weight selected from the group consisting of 48-52 Kd, 35-38 Kd and 120-135 Kd, said antigen being detectable in humans during disseminated candidiasis but not during non-invasive C. albicans infections; and
- b. detecting antibody bound by the antigen of said preparation.
- 2. A diagnostic method according to claim 1 wherein the detecting means is selected from the group consisting of latex agglutination, radioimmunoassay, enzyme-linked immunosorbent assay, and immunoblot assay.
- 3. A diagnostic method according to claim 1 wherein the antigen is the $48\text{-}52~\mathrm{Kd}$ antigen.
- 4. A diagnostic method according to claim 1 wherein the antigen is recognized by monoclonal antibody produced by a hybridoma selected from the group of hybridomas consisting of ATCC #HB-8397 and ATCC #HB-8398.
- 5. A substantially biochemically pure preparation of a <u>cytoplasmic</u> antigen of C. albicans, said antigen having an apparent molecular weight selected from the group consisting of 48-52 Kd, 35-38 Kd and 120-135 Kd, said antigen being detectable in humans during disseminated candidiasis but not during non-invansive C. albicans l infections.
- 6. A preparation according to claim 5 of the 48-52 Kd antigen.
- 7. A substantially biochemically pure preparation of a <u>cytoplasmic</u> antigen of C. albicans which is recognized by a monoclonal antibody produced by a hybridoma selected from the group of hybridomas consisting of ATCC #HB-8397 and ATCC #HB-8398.







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☐1: Infect Immun 1994 Apr;62(4):1400-5

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Production of antibodies to antigens of Candida albicans in CBA/H mice.

Costantino PJ, Franklyn KM, Gare NF, Warmington JR.

Services

School of Biomedical Sciences, Curtin University of Technology, Bentley, Perth, Australia.

Reported targets of the specific immune responses to Candida albicans in human candidiasis include a 47-kDa breakdown product of a 90-kDa heat shock protein (HSP 90) (R. Matthews and J. Burnie, FEMS Microbiol. Lett. 60:25-30, 1989) and the 48-kDa enolase (K.M. Franklyn, J.R. Warmington, A.K. Ott, and R.B. Ashman, Immunol. Cell Biol. 68:173-178, 1990). These proteins are immunodominant antigens of C. albicans. Western blotting (immunoblotting) and immunoprecipitation were used to investigate the humoral response in a mouse model of systemic candidiasis. Resolution of systemic candidiasis in CBA/H mice is associated with a high level of antibody reactivity to C. albicans antigens. A significant antibody response against a non-HSP antigen of 96 kDa which was distinct from the C. albicans HSP 90 antigen was detected. Significant antibody reactivity against an HSP of 75 kDa was also detected. We concluded that resolution of C. albicans infections in CBA/H mice was associated with antibodies to an HSP and a non-HSP of 75 and 96 kDa, respectively.

Related Resources

MeSH Terms:

- Animal
- o Antibodies, Fungal/biosynthesis*
- Antigens, Fungal/immunology*
- CD4-Positive T-Lymphocytes/immunology
- Candida albicans/immunology*
- o Female
- Heat-Shock Proteins/immunology
- Mice
- o Mice, Inbred CBA
- Precipitin Tests
- Support, Non-U.S. Gov't

Substances:

· Heat-Shock Proteins

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Production of antibodies to antigens of Candida albicans in CBA/H mice

PJ Costantino, KM Franklyn, NF Gare and JR Warmington School of Biomedical Sciences, Curtin University of Technology, Bentley, Perth, Australia.

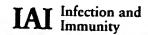
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Reported targets of the specific immune responses to Candida albicans in human candidiasis include a 47-kDa breakdown product of a 90-kDa heat shock protein (HSP 90) (R. Matthews and J. Burnie, FEMS Microbiol. Lett. 60:25-30, 1989) and the 48-kDa enolase (K.M. Franklyn, J.R. Warmington, A.K. Ott, and R.B. Ashman, Immunol. Cell Biol. 68:173-178, 1990). These proteins are immunodominant antigens of C. albicans. Western blotting (immunoblotting) and immunoprecipitation were used to investigate the humoral response in a mouse model of systemic candidiasis. Resolution of systemic candidiasis in CBA/H mice is associated with a high level of antibody reactivity to C. albicans antigens. A significant antibody response against a non-HSP antigen of 96 kDa which was distinct from the C. albicans HSP 90 antigen was detected. Significant antibody reactivity against an HSP of 75 kDa was also detected. We concluded that resolution of C. albicans infections in CBA/H mice was associated with antibodies to an HSP and a non-HSP of 75 and 96 kDa, respectively.

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- Martinez, J. P., Gil, M. L., Lopez-Ribot, J. L., Chaffin, W. L. (1998). Serologic Response to Cell Wall Mannoproteins and Proteins of Candida albicans. Clin. Microbiol. Rev. 11: 121-141
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- Chaffin, W. L., Lopez-Ribot, J. L., Casanova, M., Gozalbo, D., Martinez, J. P. (1998). Cell Wall and Secreted Proteins of Candida albicans: Identification, Function, and Expression. *Microbiol Mol Biol Rev* 62: 130-180 [Abstract] [Full Text]
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Identification of Candida albicans antigens reactive with immunoglobulin E antibody of human sera

A Ishiguro, M Homma, S Torii and K Tanaka

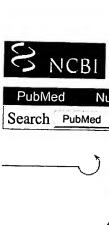
Laboratory of Medical Mycology, Nagoya University School of Medicine, Japan.

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Candida albicans antigens which reacted with immunoglobulin E (IgE) antibodies of 57 allergic patients were detected by immunoblotting. Of the various antigens, the 175-, 125-, 46-, 43-, and 37-kDa antigenic components reacted most frequently with the patient sera. To purify the major antigens, C. albicans cells were fractionated. The 46-, 43-, and 37-kDa antigens were recovered in cytoplasmic fractions, but the 175- and 125-kDa antigens were not recovered in any fraction. The 46-, 43-, and 37-kDa antigens were purified from cytoplasmic fractions by DEAE and P11 ion-exchange chromatography. Antigens were isolated by cutting bands out of sodium dodecyl sulfate-polyacrylamide gels. The purified components confirmed by immunoblotting were next processed for amino acid sequencing. Parts of the sequences of the 46-, 43-, and 37-kDa antigens had significant levels of homology with Saccharomyces cerevisiae glycolytic enzyme enolase, phosphoglycerate kinase, and aldolase, respectively. Rabbit IgG antibodies prepared against the 46- and 43-kDa antigens strongly cross-reacted with the homologous proteins of S. cerevisiae. However, S. cerevisiae enolase and phosphoglycerate kinase did not cross-react with IgE of patient sera. This result suggests that IgE antibodies against only small parts of their epitopes are elevated in the allergic patients. Since enolase is reported to be a major antigen for systemic candidiasis, this enzyme may be the immunodominant protein in both allergies and fungal infections.

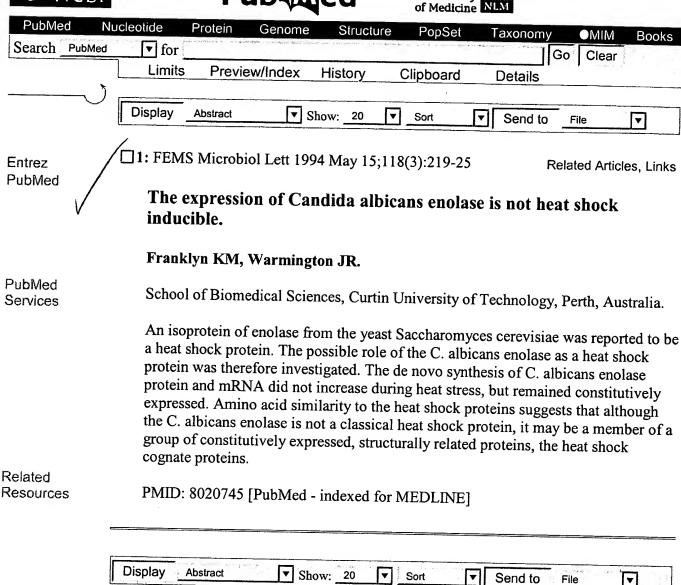
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- Gozalbo, D., Gil-Navarro, I., Azorin, I., Renau-Piqueras, J., Martinez, J. P., Gil, M. L. (1998). The Cell Wall-Associated Glyceraldehyde-3-Phosphate Dehydrogenase of Candida albicans Is Also a Fibronectin and Laminin Binding Protein. *Infect. Immun.* 66: 2052-2059 [Abstract] [Full Text]









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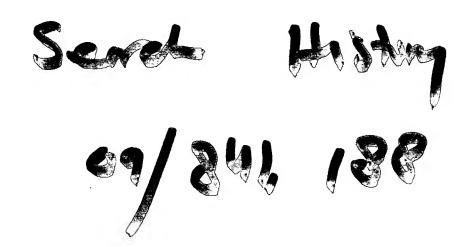
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WEST Search History

DATE: Thursday, December 12, 2002

Set Name side by side	Query	Hit Count	Set Name result set
DB = USP	PT; PLUR=YES; OP=OR		resuit set
L6	L3 and immunoassay	18	L6
L5	L3 immunoassay	18560	L5
L4	L3 and cytoplasmic	9	L4
L3	L2 and diagnosis	35	L3
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L1	((435/7.31)!.CCLS.)	169	L1

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1. 6218129. 15 May 98; 17 Apr 01. Inflammatory bowel disease first step assay system. Walsh; Michael J., et al. 435/7.21; 435/7.24 435/7.31 435/7.95 436/506 436/513. G01N033/564. 2. 6121420. 13 Apr 99; 19 Sep 00. Diagnosis of fungal infections, and a chitin-binding lectin useful in such diagnoses. Laine; Roger A., 530/350; 435/7.31. C07K014/28. 3. 5968741. 11 Apr 97; 19 Oct 99. Methods of diagnosing a medically resistant clinical subtype of ulcerative colitis. Plevy; Scott E., et al. 435/6; 435/7.31. C12Q001/68 G01N033/569. 4. 5932429. 11 Apr 97; 03 Aug 99. Methods of diagnosing clinical subtypes of crohn's disease. Targan; Stephan R., et al. 435/7.24; 435/7.31 435/7.95 435/975 436/506 436/508. G01N033/564. 5. 5914239. 08 Nov 96; 22 Jun 99. Diagnosis of fungal infections, and a chitin-binding lectin useful in such diagnoses. Laine; Roger A.. 435/7.31; 435/34 435/35 435/7.9 436/518. G01N033/53. 5707816. 06 Feb 97; 13 Jan 98. Immunological cross reactivity between Candida and human tissue or food antigens. Vojdani; Aristo. 435/7.21; 435/7.31 435/7.32 435/7.9 435/7.92 435/7.93 435/7.94 435/7.95 436/174 436/514 436/530. G01N033/53 G01N033/567 G01N033/554 G01N033/542. 4806465. 30 Mar 87; 21 Feb 89. Cytoplasmic antigens of candida albicans and methods of using the same. Buckley; Helen R., et al. 435/7.31; 435/70.21 436/518 436/548 530/371 530/388.5. G01N053/00 A61K045/00 A61K035/72 C07K015/04. 4670382. 16 Jan 84; 02 Jun 87. Monoclonal antibody to Candida albicans cytoplasmic antigens and methods of preparing same. Buckley; Helen R., et al. 435/7.31; 424/141.1 435/341 435/7.92 435/70.21 435/948 436/548 530/388.5 530/389.1. G01N033/54 C12N015/00 C12N005/00 C12R001/91. 4051232. 12 Aug 75; 27 Sep 77. Serologic test for systemic candidiasis. Protzman; Walter P., et al. 435/7.31; 435/13 436/515 436/811. G01N033/16 G01N031/02. Generate Collection Print **Terms Documents** L3 and cytoplasmic

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File: USPT

Sep 19, 2000

US-PAT-NO: 6121420

DOCUMENT-IDENTIFIER: US 6121420 A

TITLE: Diagnosis of fungal infections, and a chitin-binding lectin useful in such

DATE-ISSUED: September 19, 2000

INVENTOR-INFORMATION:

NAME

CITY

STATE

ZIP CODE

COUNTRY

Laine; Roger A.

Baton Rouge LA

US-CL-CURRENT: <u>530</u>/<u>350</u>; <u>435</u>/<u>7.31</u>

CLAIMS:

I claim:

- 1. Substantially pure chitovibrin; wherein said chitovibrin is a protein of molecular weight about 134 kDa as determined by sodium dodecyl sulfate-polyacrylamide gel electrophoresis; wherein said chitovibrin has affinity for chitin and for chito-oligomers dp9 and larger; wherein said chitovibrin is obtained from or is identical to a protein obtained from secretions from marine bacteria of the genus Vibrio induced by the presence of chitin, chitin oligomers, or cellobiose; wherein said chitovibrin has an isoelectric pH of about 3.6; wherein said chitovibrin binds chitin at an optimum pH of about 6; wherein said chitovibrin binds to chitin in aqueous solutions throughout a range of NaCl concentrations from 0 M NaCl to about 4 M NaCl; and wherein said chitovibrin has no hydrolytic activity towards chitin, towards chitin oligomers, or towards
- 2. Substantially pure chitovibrin as recited in claim 1, wherein the amino-terminal sequence of said chitovibrin is SEQ ID NO. 1.
- 3. Substantially pure chitovibrin as recited in claim 1, wherein said chitovibrin is obtained from or is identical to a protein obtained from secretions induced in the Vibrio parahemolyticus strain having accession number ATCC 27969 by the presence of chitin, chitin oligomers, or cellobiose.
- 4. A substantially pure polypeptide, wherein:
- (a) said polypeptide has affinity for chitin and for chito-oligomers dp9 and larger; and said polypeptide has no hydrolytic activity towards chitin, towards chitin oligomers, or towards chitobiose; and
- (b) said polypeptide is a fragment of chitovibrin, wherein the chitovibrin is a larger protein of molecular weight about 134 kDa as determined by sodium dodecyl sulfate-polyacrylamide gel electrophoresis; the chitovibrin has affinity for chitin and for chito-oligomers dp9 and larger; the chitovibrin is obtained from or is identical to a protein obtained from secretions from marine bacteria of the genus Vibrio induced by the presence of chitin, chitin oligomers, or cellobiose; the chitovibrin has an isoelectric pH of about 3.6; the chitovibrin binds chitin at an optimum pH of about 6; the chitovibrin binds to chitin in aqueous solutions throughout a range of NaCl concentrations from 0 M NaCl to about 4 M NaCl; and the chitovibrin has no hydrolytic activity towards chitin, towards chitin oligomers,

5. A polypeptide as recited in claim 4, wherein said polypeptide is obtained from or is identical to a protein obtained from the proteolytic breakdown of chitovibrin by endogenous protease activity of Vibrio parahemolyticus; said polypeptide has a molecular weight about 80-85 kDa as determined by sodium dodecyl sulfate-polyacrylamide gel electrophoresis; said polypeptide binds chitin at an optimum pH of about 6; said polypeptide binds to chitin in aqueous solutions throughout a range of NaCl concentrations from 0 M NaCl to about 4 M NaCl.

L4: Entry 5 of 9

File: USPT

Jun 22, 1999

US-PAT-NO: 5914239

DOCUMENT-IDENTIFIER: US 5914239 A

TITLE: <u>Diagnosis</u> of fungal infections, and a chitin-binding lectin useful in such <u>diagnoses</u>

DATE-ISSUED: June 22, 1999

INVENTOR - INFORMATION:

NAME

CITY

STATE ZIP CODE

COUNTRY

Laine; Roger A.

Baton Rouge

LA

US-CL-CURRENT: 435/7.31; 435/34, 435/35, 435/7.9, 436/518

CLAIMS:

I claim:

- 1. A method for detecting chitin in a sample, comprising the steps of:
- (a) contacting the sample with a substance comprising chitovibrin; and
- (b) inspecting the sample for the presence of chitovibrin bound to chitin, wherein bound chitovibrin indicates the presence of chitin in the sample;

wherein said chitovibrin is a protein of molecular weight about 134 kDa as determined by sodium dodecyl sulfate-polyacrylamide gel electrophoresis; wherein said chitovibrin has affinity for chitin and for chito-oligomers dp9 and larger; wherein said chitovibrin is obtained from or is identical to a protein obtained from secretions from marine bacteria of the genus Vibrio induced by the presence of chitin, chitin oligomers, or cellobiose; wherein said chitovibrin has an isoelectric pH of about 3.6; wherein said chitovibrin binds chitin at an optimum pH of about 6; wherein said chitovibrin binds to chitin in aqueous solutions throughout a range of NaCl concentrations from 0 M NaCl to about 4 M NaCl; and wherein said chitovibrin has no hydrolytic activity towards chitin, towards chitin oligomers, or towards chitobiose.

- 2. A method as recited in claim 1, wherein said chitovibrin is conjugated to a detectable label.
- 3. A method as recited in claim 2, wherein the detectable label is selected from the group consisting of a radioactive material, a fluorophore, a dye, an electron-dense compound, and an enzyme.
- 4. A method as recited in claim 1, wherein the sample comprises a plant tissue, an agricultural product, an animal tissue, a human tissue, a contact lens, a prosthetic device, or an air filter.
- 5. A method as recited in claim 4, wherein said inspecting step comprises inspecting the sample for the presence of chitovibrin bound to chitin in fungal cell walls.
- 6. A method as recited in claim 4, wherein said inspecting step comprises inspecting the sample for the presence of chitovibrin bound to chitin in yeast bud scars.
- 7. A method as recited in claim 1, wherein the sample comprises an animal body

- fluid, a human body fluid, a plant fluid, potable water, or a beverage.
- 8. A method as recited in claim 7, wherein said inspecting step comprises inspecting the sample for the presence of chitovibrin bound to chitin in fungal cell walls.
 - 9. A method as recited in claim 7, wherein said inspecting step comprises inspecting the sample for the presence of chitovibrin bound to chitin in yeast bud scars.
 - 10. A method as recited in claim 1, wherein said contacting step additionally comprises contacting the sample with a reagent comprising an antibody to chitovibrin.
 - 11. A method as recited in claim 10, wherein said reagent additionally comprises a detectable label.
 - 12. A method as recited in claim 11, wherein the detectable label is selected from the group consisting of a radioactive material, a fluorophore, a dye, an electron-dense compound, and an enzyme.
 - 13. A method as recited in claim 10, wherein the sample comprises a plant tissue, an agricultural product, an animal tissue, a human tissue, a contact lens, a prosthetic device, or an air filter.
 - 14. A method as recited in claim 13, wherein said inspecting step comprises inspecting the sample for the presence of chitovibrin bound to chitin in fungal cell walls.
 - 15. A method as recited in claim 13, wherein said inspecting step comprises inspecting the sample for the presence of chitovibrin bound to chitin in yeast bud
 - 16. A method as recited in claim 10, wherein the sample comprises an animal body fluid, a human body fluid, a plant fluid, potable water, or a beverage.
 - 17. A method as recited in claim 16, wherein said inspecting step comprises inspecting the sample for the presence of chitovibrin bound to chitin in fungal cell walls.
- 18. A method as recited in claim 16, wherein said inspecting step comprises inspecting the sample for the presence of chitovibrin bound to chitin in yeast bud scars.
- 19. A method for detecting chitin in a sample, comprising the steps of contacting the sample with a substance comprising a polypeptide, and inspecting the sample for the presence of said polypeptide bound to chitin, wherein bound polypeptide indicates the presence of chitin in the sample; wherein:
- (a) said polypeptide has affinity for chitin and for chito-oligomers dp9 and larger; and said polypeptide has no hydrolytic activity towards chitin, towards chitin oligomers, or towards chitobiose; and
- (b) said polypeptide is obtained from or is identical to a polypeptide obtained from the proteolytic breakdown of chitovibrin by endogenous protease activity of Vibrio parahemnolyticus; said polypeptide has a molecular weight about 80-85 kDa as determined by sodium dodecyl sulfate-polyacrylamide gel electrophoresis; said polypeptide binds chitin at an optimum pH of about 6; said polypeptide binds to chitin in aqueous solutions throughout a range of NaCl concentrations from 0 M NaCl to about 4 M NaCl; wherein chitovibrin is a larger protein of molecular weight about 134 kDa as determined by sodium dodecyl sulfate-polyacrylamide gel electrophoresis; wherein chitovibrin has affinity for chitin and for chito-oligomers dp9 and larger; wherein chitovibrin is obtained from or is identical to a protein obtained from secretions from marine bacteria of the genus Vibrio induced by the presence of chitin, chitin oligomers, or cellobiose; wherein

chitovibrin has an isoelectric pH of about 3.6; wherein chitovibrin binds chitin at an optimum pH of about 6; wherein chitovibrin binds to chitin in aqueous solutions throughout a range of NaCl concentrations from 0 M NaCl to about 4 M NaCl; and

wherein chitovibrin has no hydrolytic activity towards chitin, towards chitin oligomers, or towards chitobiose.

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- 2. <u>6300065</u>. 26 Aug 98; 09 Oct 01. Yeast cell surface display of proteins and uses thereof. Kieke; Michele C., et al. 435/6; 435/252.3 435/254.1 435/254.11 435/254.21 435/254.22 435/254.23 435/29 435/320.1 435/325 435/348 435/69.1 435/7.1 435/7.2 <u>435/7.31</u>. C12P021/02 C12Q001/68 C12Q001/02 C12N015/63 C12N015/81.
- 3. <u>6218129</u>. 15 May 98; 17 Apr 01. Inflammatory bowel disease first step assay system. Walsh; Michael J., et al. 435/7.21; 435/7.24 <u>435/7.31</u> 435/7.95 436/506 436/513. G01N033/564.
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- 6. <u>5968741</u>. 11 Apr 97; 19 Oct 99. Methods of diagnosing a medically resistant clinical subtype of ulcerative colitis. Plevy; Scott E., et al. 435/6; <u>435/7.31</u>. C12Q001/68 G01N033/569.
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- 10. <u>5541077</u>. 13 Dec 94; 30 Jul 96. Fungal stress proteins. Burnie; James P., et al. <u>435/7.31</u>; 435/7.92 435/7.95 436/530 436/534 436/815 530/387.9 530/388.5 530/389.1. G01N033/569 C07K016/14.

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- 11. 5187064. 20 Dec 88; 16 Feb 93. Monoclonal antibodies and methods for fungal pathogen detection. Petersen; Frank P., et al. 435/7.31; 435/34 435/341 435/7.9 436/518 436/548 530/388.5. C07K015/02 C12Q001/04 G01N033/577. 12. 5043267. 16 Jan 90; 27 Aug 91. Method for rapid detection of bacterial and fungal infection. Richards; James C.. 435/7.31; 435/7.32 435/7.33 435/961 436/17 436/174 436/177 436/519 436/522.
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- 17. 4670382. 16 Jan 84; 02 Jun 87. Monoclonal antibody to Candida albicans cytoplasmic antigens and methods of preparing same. Buckley; Helen R., et al. 435/7.31; 424/141.1 435/341 435/7.92 435/70.21 435/948 436/548 530/388.5 530/389.1. G01N033/54 C12N015/00 C12N005/00 C12R001/91.
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☐ 14. <u>5932429</u> . 11 Apr 97; 03 Aug 99. Methods of di Targan; Stephan R., et al. 435/7.24; <u>435/7.31</u> 435/7.95 435	/975 436/506 436/508. G01N033/564.	
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